

## Abstract

The invention provides a method for analyzing the hydrogen isotopes, in particular, a method for confirming hydrogen/deuterium exchange in the structure of a biological sample and determining the percent content of deuterium in a biological sample. The method comprises the following steps: selecting a matrix, mixing homogeneously the prepared biological sample with a suitable amount of matrix followed by lyophilization, sufficiently oxidizing the lyophilized mixed sample with oxidants through burning in order to oxidize the hydrogen in the mixed sample to water, reacting the resulting water after separation with zinc to generate hydrogen gas, determining the  $^2\text{H}/^1\text{H}$  ratio of the hydrogen gas with a gas isotope mass spectrometer, and then calculating the percent content of deuterium in the biological sample through a formula. When the percent content of deuterium in a biological sample increases, its biological characters are markedly changed, and mainly, the heat resistance is markedly changed. The percent content of deuterium in a biological sample having optimal heat stability and the optimal deuteration conditions of a biological sample are ascertained, and the stability of biological sample such as polypeptide is improved. This method has a good repeatability of determination, is simple and easy to manipulate, requires a small amount of sample used, and has low cost.